

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listings, of claims in the application:

**Listing of Claims:**

1. (Original) A method of stimulating the immune system of a subject which comprises administering to the subject an immunologically effective amount of an immunostimulatory molecule which comprises at least one oligonucleotide strand which comprises

(1) at least one nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleobase, and thereby also comprising at least one CxG dinucleotide unit or analogue thereof, and

(2) at least one covalently incorporated lipophilic group.

2. (Original) The method of claim 1 in which element (1) comprises a CxG dinucleotide unit.

3. (Currently Amended) The method of ~~claims 1 or 2~~  
claim 1 in which the CxG dinucleotide unit is a CpG  
dinucleotide unit.

4. (Currently Amended) The method of ~~any one of~~  
~~claims 1-3~~ claim 1 in which at least one lipophilic group is a  
strongly lipophilic group.

5. (Currently Amended) The method of ~~any one of~~  
~~claims 1-4~~ claim 1 in which at least one lipophilic group is a  
highly lipophilic (Meylan) group.

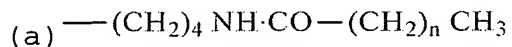
6. (Currently Amended) The method of ~~any one of~~  
~~claims 1-5~~ claim 1 in which at least one lipophilic group has  
a predicted logP, according to the Meylan algorithm, of at  
least 4.

7. (Currently Amended) The method of ~~any one of~~  
~~claims 1-5~~ claim 1 in which at least one lipophilic group has  
a predicted logP, according to the Meylan algorithm, of at  
least 7.

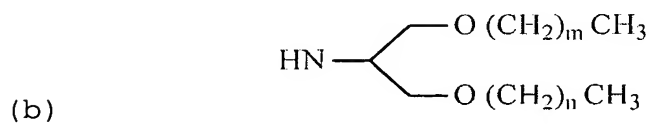
8. (Currently Amended) The method of ~~any one of~~  
~~claims 1-5~~ claim 1 in which at least one lipophilic group has

a predicted logP, according to the Meylan algorithm, of at least 10.

9. (Currently Amended) The method of ~~any one of claims 1-8~~ claim 1 in which at least one lipophilic group is selected from the group consisting of



where  $n$  = an integer with values ranging from 6 to 26,



where  $m$  and  $n$  are independent integers with values ranging from 6 to 26,

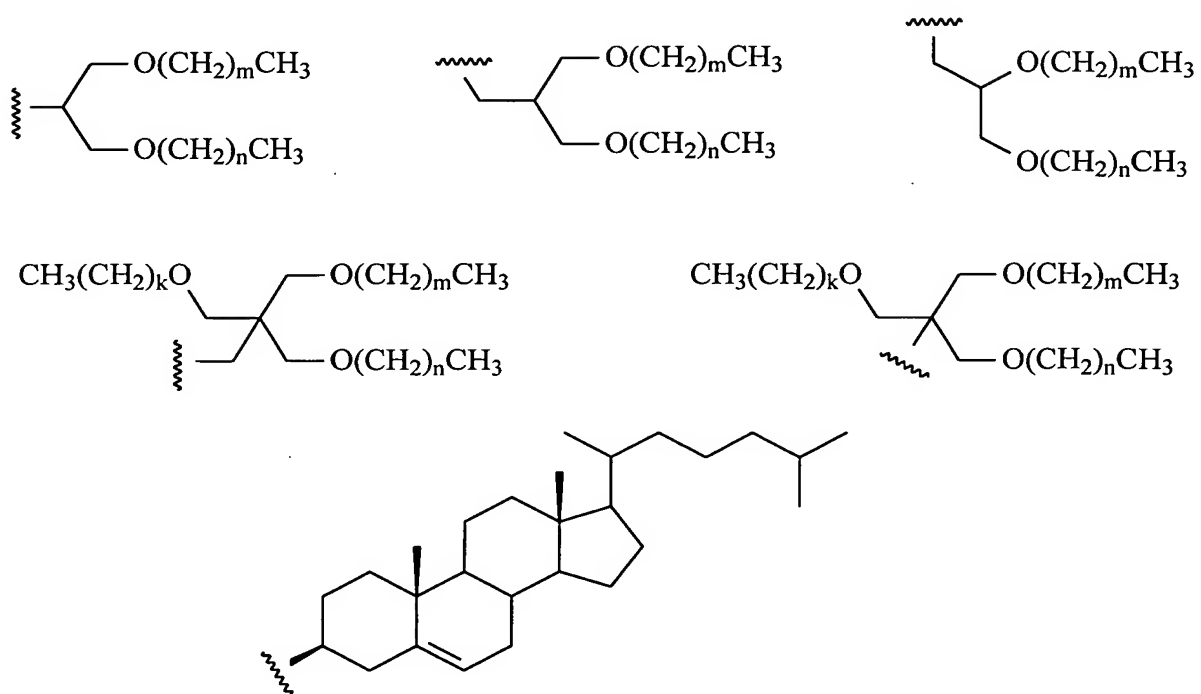
(c)

-XR wherein X is -O-, -S-, or -NH- and -R is aliphatic, and

(d)

-XR wherein X is -O-, -S-, or -NH- and -R is at least partially aromatic[[],]\_.

10. (Original) The method of claim 9 where -R is a group selected from the group consisting of one of the following structures:



where m, n, and k are independent integers with values ranging from 3 to 30.

11. (Currently Amended) The method of ~~any one of claims 1-9~~ claim 1 in which at least one lipophilic group is one of the lipophilic groups depicted in Fig. 2.

12. (Currently Amended) The method of ~~any one of claims 1-11~~ claim 1 where said molecule comprises at least two lipophilic groups.

13. (Original) The method of claim 12 where said molecule comprises at least two strongly lipophilic groups.

14. (Original) The method of claim 12 where said molecule comprises at least two highly lipophilic (Meylan) groups.

15. (Currently Amended) The method of ~~any one of claims 1-14~~ claim 1 in which there are fewer than 8 nucleobases on each nucleotide strand.

16. (Original) The method of claim 15 in which there are fewer than 5 nucleobases on each nucleotide strand.

17. (Currently Amended) The method of ~~any one of claims 1-16~~ claim 1 in which each of the nucleobases is selected from the group consisting of adenine, guanine, thymine, cytosine, uracil, and hypoxanthine.

18. (Currently Amended) The method of ~~any one of claims 1-16~~ claim 1 in which each of the nucleobases is selected from the group consisting of adenine, guanine, thymine, and cytosine.

19. (Currently Amended) The method of ~~any one of claims 1-18~~ claim 1 wherein at least one nucleotide comprises a nucleobase-carbohydrate nucleoside.

20. (Original) The method of claim 19 in which the carbohydrate is a monosaccharide.

21. (Original) The method of claim 19 in which the number of carbon atoms in the monosaccharide is 3-8.

22. (Original) The method of claim 21 in which the monosaccharide is a pentose.

23. (Currently Amended) The method of ~~any one of claims 20-22~~ claim 20 in which the monosaccharide is an aldose.

24. (Currently Amended) The method of ~~any one of claims 20-23~~ claim 20 in which the monosaccharide is cyclized.

25. (Original) The method of claim 19 in which the monosaccharide is a pyranose.

26. (Original) The method of claim 20 in which the monosaccharide is a ribose or a 2-deoxyribose.

27. (Currently Amended) The method of ~~any one of claims 1-26~~ claim 1 in which all of the nucleotides of one strand comprise a nucleobase-carbohydrate nucleoside.

28. (Currently Amended) The method of ~~any one of claims 1-27~~ claim 1 in which at least one nucleotide further comprises a phosphate.

29. (Original) The method of claim 28 in which at least one nucleotide comprises one and only one phosphate.

30. (Original) The method of claim 29 in which at least one strand is DNA or RNA.

31. (Currently Amended) The method of ~~any one of claims 1-30~~ claim 1 in which at least one lipophilic group is covalently incorporated into a free end of at least one strand.

32. (Currently Amended) The method of ~~any one of claims 1-31~~ claim 1 in which at least one lipophilic group is covalently incorporated into the 3' end of at least one strand.

33. (Currently Amended) The method of ~~claims 31 or 32~~ claim 31 in which the lipophilic group is attached to the end through a phosphate group.

34. (Currently Amended) The method of ~~any one of claims 1-33~~ claim 1 in which at least one lipophilic group is incorporated into an internucleoside linkage.

35. (Currently Amended) The method of ~~any one of claims 1-34~~ claim 1 in which at least one lipophilic group is a substituent of a nucleobase.



36. (Currently Amended) The method of ~~any one of claims 1-35~~ claim 1 in which at least part of the oligonucleotide has a backbone which differs from that of DNA and RNA.

37. (Original) The method of claim 36 in which the backbone differs in that the internucleoside linkage is not a phosphate group.

38. (Original) The method of claim 36 in which the backbone differs in that at least one nucleotide is a non-normal nucleotide which does not comprise ribose or 2-deoxyribose.

39. (Original) The method of claim 38 in which the non-normal nucleoside comprises a sugar.

40. (Original) The method of claim 38 in which the non-normal nucleotide does not comprise a sugar.

41. (Original) The method of claim 40 in which the oligonucleotide is at least partially a PNA oligomer.

42. (Original) The method of claim 40 in which at least one non-normal nucleotide comprises a non-normal nucleoside of the form

nucleobase-O-alkyl

where the -O-alkyl is the residue of a polyol, and the alkyl is not more than 6 carbon atoms.

43. (Original) The method of claim 42 wherein the polyol is glycerol, and hence the alkyl is 3 carbon atoms.

44. (Currently Amended) The method of ~~any one of claims 36-40, 42 or 43~~ claim 36 in which at least non-normal nucleoside is bound to a phosphate group.

45. (Original) The method of claim 44 in which there are two adjacent such non-normal nucleosides and the internucleoside linkage between them is a phosphate group.

46. (Original) The method of claim 44 in which there are two adjacent such non-normal nucleosides and the internucleoside linkage between them is

In re of: 10/502,085 JIANG4A

-phosphate group-linker Z-phosphate group-,

where linker Z is aliphatic.

47. (Original) The method of claim 46 in which linker Z is of the form  $-\text{[small alkyl-O]}_n$ , where n is 1 to 20, and small alkyl is not more than 6 carbon.

48. (Original) The method of claim 47 in which linker Z is  $-\text{[CH}_2\text{CH}_2\text{O]}_n-$ .

49. (Currently Amended) The method of ~~any one of claims 1-49~~ claim 1 in which the dinucleotide unit comprises a non-natural nucleoside, or an internucleoside linkage which is not a phosphate group.

50. (Currently Amended) The method of ~~any one of claims 1-48~~ claim 1 in which the CxG dinucleotide unit comprises two non-natural nucleosides and the internucleoside linkage between them is a phosphate group.

51. (Original) The method of claim 49 in which the dinucleotide unit comprises two non-natural nucleosides and the internucleoside linkage between them is

In re of: 10/502,085 JIANG4A

-phosphate group-linker Z-phosphate group-,

where linker Z is aliphatic.

52. (Original) The method of claim 51 in which linker Z is  $-\text{[CH}_2\text{CH}_2\text{O]}_n-$  and n is 1 to 20.

53. (Original) The method of claim 49 in which the dinucleotide unit is a PNA oligomer.

54. (Original) The method of claim 49 in which the dinucleotide unit is a GNA oligomer.

55. (Currently Amended) The method of ~~any one of claims 1-54~~ claim 1 in which the molecule lacks double stranded structure.

56. (Currently Amended) The method of ~~any one of claims 1-54~~ claim 1 in which the molecule has at least some double stranded structure.

57. (Currently Amended) The method of ~~any one of claims 1-56~~ claim 1 in which there are no more than seven nucleobases in each oligonucleotide strand.

58. (Currently Amended) The method of ~~any one of claims 1-56~~ claim 1 in which there are no more than four nucleobases in each oligonucleotide strand.

59. (Currently Amended) The method of ~~any one of claims 1-58~~ claim 1 in which the molecule further comprises at least one epitope.

60. (Original) The method of claim 59 wherein at least one epitope is a carbohydrate epitope.

61. (Currently Amended) The method of claim 59 ~~or 60~~ wherein at least one epitope is a peptide epitope.

62. (Currently Amended) The method of ~~any one of claims 59-61~~ claim 59 wherein at least one epitope is a B-cell epitope.

63. (Currently Amended) The method of ~~any one of claims 59-62~~ claim 59 wherein at least one epitope is a T-cell epitope.

64. (Currently Amended) The method of ~~any one of claims 59-63~~ claim 59 wherein at least one epitope is a MUC1 epitope.

65. (Currently Amended) The method of ~~any one of claims 1-64~~ claim 1 in which the oligonucleotide is cyclized, so as to lack a free end, and the lipophilic groups are incorporated elsewhere in the molecule.

66. (Currently Amended) The method of ~~any one of claims 1-64~~ claim 1 where said molecule which comprises two or more segments, each segment consisting of nucleosides joined to each other by short internucleoside linkages, each segment being joined to at least one other segment by a long internucleoside linkage,

at least two of said segments each comprising at least one CxG dinucleotide unit or analogue thereof.

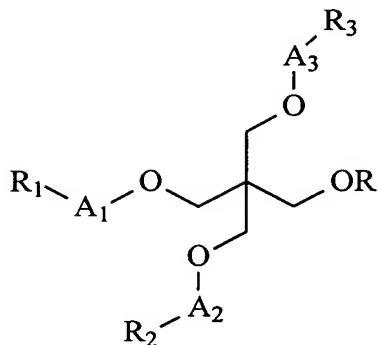
67. (Original) The method of claim 66 in which said segments are connected by said internucleoside linkages to form one or more linear chains.

68. (Original) The method of claim 66 in which two or more segments are cyclized by two or more internucleoside linkages.

69. (Original) The molecule of claim 66 in which at least one internucleoside linkage is a higher order linkage which simultaneously links three or more segments to each other, whereby said segments are connected to form a branched structure.

70. (Original) The molecule of claim 66 in which at least one internucleoside linkage comprises a pentaerythritol unit.

71. (Original) The molecule of claim 66 in which the molecule has the following structural feature



wherein R is a lipophilic group;  $A_1$ ,  $A_2$  and  $A_3$  are independently a linking arm of 0 to 20 atoms length;  $R_1$ ,  $R_2$  and  $R_3$  are independently chosen from the group consisting of a hydrogen atom, a lipophilic group or an oligo-nucleotide residue.

72. (Currently Amended) The method of ~~any one of claims 1-71~~ claim 1 where said nucleotide sequence comprises at least one pair of adjacent thymine nucleobases which are dimerized to form a thymine dimer.

73. (Currently Amended) The method of ~~any one of claims 1-72~~ claim 1 in which at least one lipophilic group is



covalently incorporated into the 5' end of an oligonucleotide strand.

74. (Currently Amended) The method of ~~any one of claims 1-73~~ claim 1 in which the molecule does not have cytotoxic activity against cancer cells.

75. (Currently Amended) The method of ~~any one of claims 1-74~~ claim 1 in which the subject is not suffering from a cancer.

76. (Currently Amended) The method of ~~any one of claims 1-75~~ claim 1 in which the subject is not being medicated with any otehr cancer preventative.

77. (Currently Amended) The method of ~~any one of claims 1-76~~ claim 1 in which the molecule potentiates the specific innate immune response to a pathogen or cancer already present in the subject.

78. (Currently Amended) The method of ~~any one of claims 1-76~~ claim 1 which further comprises administering a pharmaceutical composition comprising an immunogen to the

subject, said molecule potentiating the specific elicited immune response to said immunogen.

79. (Original) The method of claim 78 in which the molecule and the immunogen are administered simultaneously.

80. (Original) The method of claim 79 in which the molecule and the immunogen are administered in the same composition.

81. (Original) The method of claim 78 in which the immunostimulatory oligonucleotide molecule is also an immunogen which elicits a specific immune response protective against said pathogen or cancer.

82. (Cancelled)

83. (Original) An immunostimulatory molecule which comprises at least one oligonucleotide strand, said strand comprising a nucleotide sequence, the nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleobase, and thereby also comprising at least one CxG dinucleotide unit or analogue thereof, and

(2) at least one covalently incorporated lipophilic group,

where the number of nucleotides in said strand is less than eight.

84. (Currently Amended) The molecule of claim 83 which comprises at least one oligonucleotide strand, said strand comprising a nucleotide sequence, the nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleobase, and thereby also comprising at least one CxG dinucleotide unit, and

(2) at least one covalently incorporated lipophilic group, where the number of nucleotides in said strand is less than eight ~~where the nature of said molecule is further limited in accordance with any one of claims 2-25.~~

85. (Original) An immunostimulatory molecule which comprises (I) at least one oligonucleotide strand which comprises

(1) at least one nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleobase, and thereby also comprising at least one CxG dinucleotide unit or analogue thereof, and

(2) at least one covalently incorporated lipophilic group, and

(II) an epitope.

86. (Original) The molecule of claim 85 wherein at least one epitope is a carbohydrate epitope.

87. (Currently Amended) The molecule of claim 85 ~~or 86~~ wherein at least one epitope is a peptide epitope.

88. (Currently Amended) The molecule of ~~any one of claims 85-87~~ claim 85 wherein at least one epitope is a B-cell epitope.

89. (Currently Amended) The molecule of ~~any one of claims 85-88~~ claim 85 wherein at least one epitope is a T-cell epitope.

90. (Currently Amended) The molecule of ~~any one of claims 85-89~~ claim 85 wherein at least one epitope is a MUC1 epitope.

91. (Cancelled)

92. (Original) An immunostimulatory molecule which comprises (I) at least one oligonucleotide strand which comprises

(1) at least one nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleoside, each nucleoside comprising a nucleobase, each nucleoside being joined to at least one other nucleoside by an internucleoside linkage,

said molecule comprising two or more segments, each segment consisting of nucleosides joined to each other by short internucleoside linkages, each segment being joined to at least one other segment by a long internucleoside linkage,

at least two of said segments each comprising at least one CxG dinucleotide unit or analogue thereof, and

(2) at least one covalently incorporated lipophilic group.

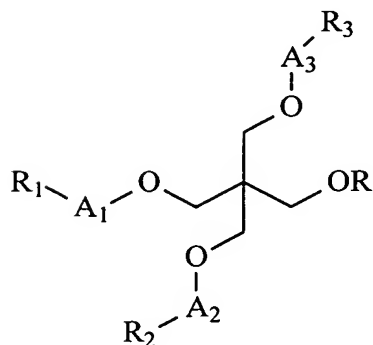
93. (Original) The molecule of claim 92 in which said segments are connected by said internucleoside linkages to form one or more linear chains.

94. (Original) The molecule of claim 92 in which one or more segments are connected by one or more of said long internucleoside linkages to form a cyclic structure.

95. (Original) The molecule of claim 92 in which at least one internucleoside linkage is a higher order linkage which simultaneously links three or more segments to each other, whereby said segments are connected to form a branched structure.

96. (Original) The molecule of claim 92 in which at least one internucleoside linkage comprises a pentaerythritol unit

97. (Original) The molecule of claim 92 in which the molecule has the following structural feature



wherein R is a lipophilic group; A<sub>1</sub>, A<sub>2</sub> and A<sub>3</sub> are independently a linking arm of 0 to 20 atoms length; R<sub>1</sub>, R<sub>2</sub> and R<sub>3</sub> are independently chosen from the group consisting of a hydrogen atom, a lipophilic group or an oligo-nucleotide residue.

98. (Currently Amended) The molecule of ~~any one of claims 92-97~~ claim 92 which comprises (I) at least one oligonucleotide strand which comprises

(1) at least one nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleoside, each nucleoside comprising a nucleobase, each nucleoside being joined to at least one other nucleoside by an internucleoside linkage,

said molecule comprising two or more segments, each segment consisting of nucleosides joined to each other by short internucleoside linkages, each segment being joined to at least one other segment by a long internucleoside linkage,

at least two of said segments each comprising at least one CxG dinucleotide unit, and (2) at least one covalently incorporated lipophilic group ~~where the nature of the molecule is further limited in accordance with any one of claims 2-64.~~

99. (Original) An immunostimulatory molecule which comprises at least one oligonucleotide strand, said strand comprising

(1) a nucleotide sequence, the nucleotide sequence comprising a plurality of nucleotides, each nucleotide comprising a nucleobase, and thereby also comprising at least one CxG dinucleotide unit or analogue thereof, and

(2) at least one covalently incorporated lipophilic group,

where said nucleotide sequence comprises at least one pair of adjacent thymine nucleobases which are dimerized to form a thymine dimer.

100. (Currently Amended) The method of ~~any one of claims 1-81~~ claim 1 where none of the internucleoside linkages is selected from the group consisting of poly(N-vinyl), poly(methacryloxyethyl), poly(methacrylamide), and poly(etheylenimine).